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EFFECTS OF EYESTALK ABLATION ON LARVAL MOLTING RATES AND MORPHOLOGICAL DEVELOPMENT OF THE AMERICAN LOBSTER, *HOMARUS AMERICANUS*

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Abstract

Larvae of the lobster, *Homarus americanus*, displayed abbreviation of subsequent molt intervals in response to bilateral eyestalk ablation at the 2nd stage. The interval in which the ablation occurred (2nd stage) was occasionally shortened. However, the 3rd stage molt interval was always abbreviated. At the 4th stage the ablated larvae were larger than intact controls. Removal of the last pair of swimming legs, as a control for the trauma of eyestalk ablation, slightly decreased growth but had no effect on molt interval lengths. Eyestalk removal sometimes resulted in 4th stage larvae with characteristics intermediate between normal 3rd and 4th stages. Eyestalkless larvae that were reared communally and fed frozen *Artemia* formed significant numbers of these intermediates. Eyestalkless larvae reared individually and fed live *Artemia* nauplii rarely formed intermediates. These results imply that although eyestalks may be involved in the regulation of both the timing and growth increment of larval molting, they may not mediate the morphological aspects of lobster metamorphosis.

Introduction

Crustacean growth involves the interrelationship of two components. The first is the molt interval. Its regulation by molt-inhibiting hormone and ecdysteroids has been reviewed recently by Skinner (1985). The other component of growth is the molt increment, which is the increase in size that occurs from one stage to the next. Size increase occurs by absorption of water leading to an expansion of the new, soft exoskeleton and by slight stretching of the arthrodial membranes during intermolt (see Hartnoll, 1982, for review).

Development for many crustacean larvae consists of gradual morphological changes with successive molts until the first postlarval (juvenile) stage. In many instances, the gradual changes can be likened to those occurring in the hemimetabolous insects (Granger and Bollenbacher, 1981, for review). This process of gradual but pronounced changes through larval development can be considered a true metamorphosis (Passano, 1961). Usually, a standard number of stages is observed in laboratory rearing studies, each with its characteristic growth increment (Williamson, 1982). Variable numbers of stages, however, can also occur (Costlow, 1965; Williamson, 1982).

Marine crustacean larvae are usually tolerant of a wide range of temperature and salinity regimes. Suboptimal conditions can result in larvae that display extra intermediate stages (possessing characteristics of the normal preceding and subsequent stages) before completion of metamorphic development (Boyd and Johnson, 1963; Knowlton, 1974). Knowlton (1974) considered a variety of factors and reasoned that

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temperature and nutritional state were generally the most important in larval development.

Larval intermediate stages have also resulted from eyestalk ablation during zoeal development. The suggestion has been made that eyestalks are involved in the control of metamorphosis (Costlow, 1966b, 1968; Little, 1969; Charmantier *et al.*, 1985). Extra zoeal stages were observed after destalking the crabs *Sesarma reticulatum* (Costlow, 1966a) and *Rhithropanopeus harrisii* (Costlow, 1966b), and the shrimp *Palaemon macrodactylus* (Little, 1969). Intermediate stages in the lobster *Homarus americanus* were recently observed following eyestalk removal (Charmantier *et al.*, 1985) similar to those seen occasionally in normal laboratory-reared larvae (Templeman, 1936).

This paper presents additional data on the various effects of eyestalk ablation and rearing conditions on larval molting and development of *Homarus americanus*. We show that eyestalk ablation abbreviates subsequent larval molt intervals. We also provide evidence that suggests that eyestalks may not be involved in the mediation of morphological development of larval lobsters.

Materials and Methods

Experimental animals and maintenance

American lobsters, *Homarus americanus*, used in these studies were from a variety of sources. Adult, egg-bearing females were kindly supplied by Dr. John Castell, Halifax Laboratory, Nova Scotia. Others were shipped from the Massachusetts State Lobster Hatchery, Martha's Vineyard, as gifts from John Hughes. Some of the females were cultured, and the larvae obtained at the Bodega Marine Laboratory by established techniques (Chang and Conklin, 1983; Hedgecock, 1983). Larvae were removed from the hatch system (18°C) within 6 h of hatching, placed into 4-liter beakers containing aerated, 1 μm-filtered, UV-irradiated seawater, and allowed to acclimate to the temperature at which they were subsequently raised (18–20°C, unless otherwise noted). For a single experiment, larvae from a single day's hatch were routinely used.

Larvae were individually transferred after acclimation to randomly assigned single compartments of plastic parts trays (Grainger's #A824). Compartments were filled with 50 ml of 1 μm-filtered, UV-irradiated seawater containing 12.5 units/ml of penicillin, 12.5 μg/ml of streptomycin, and 31.3 ng/ml of fungizone (antibiotic mix, Gibco). These concentrations are well within those used in other studies (Brick, 1974; Fisher and Nelson, 1977). As in those studies, the only observable effect of antibiotics on larvae was to prevent microbial infestations and thereby increase survival. *Artemia* nauplii, freshly hatched by the decapsulation method (Sorgeloos *et al.*, 1977), were fed daily to give a final concentration of 10/ml in each compartment. At this concentration, lobster larvae were fed to satiation as indicated by large numbers of *Artemia* nauplii remaining 24 h later. Equivalent concentrations of nauplii were given with each daily seawater (with antibiotics) change.

Molting experiments

In the first experiment, lobster larvae were transferred into plastic trays as outlined above. They were observed 3× daily for the appearance of newly molted 2nd stage zoea. This resulted in larvae that could be grouped within 8 h of their previous molt. Larvae were thus eyestalk-ablated at 24 ± 4 h after the molt. Larvae were randomly selected as intact controls and for eyestalk ablation or removal of the last pair of thoracopods (swimming legs). Larvae to be ablated were individually placed on a depression slide. The surrounding seawater was removed, and both eyestalks or thor-

acopods were severed at their bases using a pair of fine scissors (Vannas, Fine Science Tools). Larvae were allowed to remain on the slide for approximately 15 s for initial blood clotting before placement into fresh seawater. Controls were placed on depression slides without seawater for an equivalent amount of time. Larvae were observed 3× daily to determine the mean molting intervals from 2nd to 3rd (2nd stage molt interval) and 3rd to 4th (3rd stage molt interval) stages. For the purposes of this work, the hatching molt stage from prelarva to the 1st stage lobster was not considered as the 1st molt interval (Davis, 1964; Aiken, 1980). Survival of ablated larvae was typically 95% or better under these conditions. Statistical significance was determined by Student *t*-tests.

Morphological experiments

Sibling larvae from a single day's hatch were placed into compartments as stated previously. They were reared throughout this experiment at 22–23°C and observed for molting to 2nd stage every 2 h. At specified times of 1, 3, 6, 12, 18, 24, 30, 36, 42, or 48 h after molting, larvae were destalked or left intact to serve as controls. Larvae were observed 3×/day for determination of the 3rd stage molt interval. Morphological measurements of the larvae were taken after the 3rd molt. These measurements consisted of antenna length from the base, carapace length from the posterior base of the eyesocket to the anterior edge of the first abdominal segment, and length of the cheliped propopodite (hereafter referred to as chela length). All measurements were made with the aid of a dissecting microscope (Wild M5) equipped with an ocular micrometer. Statistical significance of morphological and molt interval differences between groups was tested by ANOVA and Scheffe tests.

A second experiment was designed to determine the effect of rearing conditions on the development and molting of larvae that were destalked at various times. Upon hatching, larvae from a single day's hatch from another female lobster were placed into a larval rearing kreisel at 18°C (Chang and Conklin, 1983). Larvae were initially stocked at a density of 500/40-liter system, fed frozen *Artemia* (San Francisco Bay Brand) *ad libitum*, and checked 3×/day for the appearance of newly molted 2nd stage zoea. Larvae were removed after molt to 2nd stage, and were either reared individually in compartments as before (with *Artemia* nauplii) or left in the communal rearing kreisels. At 18 ± 4 h and 90 ± 4 h after the molt to 2nd stage, a portion of the kreisel- and compartment-reared larvae were destalked and returned to their respective rearing systems. Larvae were checked several times daily to determine the numbers molting to intermediate stages at the molt to 4th stage. The experiment was terminated before any of the larvae had completed the molt to 5th stage.

Results

Molting experiments

In seven of nine replicate experiments, the 2nd stage molt interval was shortened by eyestalk ablation. In a typical experiment, intact lobsters had an interval of 4.3 ± 0.2 days, whereas eyestalk-ablated larvae had an interval of 4.1 ± 0.1 days ($n = 74$, $P < 0.001$). In contrast to the occasional (78% of the experiments) abbreviation of the 2nd stage molt interval, eyestalk ablation (during early 2nd stage) always shortened the 3rd and subsequent stages (Table I). To determine whether the trauma of eyestalk removal itself was affecting subsequent molts, an additional group of 2nd stage larvae underwent removal of the last pair of thoracopods. The results in Table I indicate that removal of swimming legs had no effect on the subsequent 3rd stage molt interval.

TABLE I

Third stage molt intervals of eyestalk-ablated, leg-ablated, and intact control groups of larval lobsters

Treatment	3rd stage molt interval (days)		
	Mean	S.D.	n
Intact controls	6.2	0.5	28
Eye-ablated	5.4[a]	0.7	31
Leg-ablated	5.9[b]	0.6	22

[a] Denotes significance from the control group ($P < 0.01$, other experiments $P < 0.001$).
[b] Denotes significance from eyestalk-ablated group ($P < 0.05$) and lack of significance from controls ($P > 0.05$).

Morphological experiments

The most apparent effect of ablation at various times after the molt to 2nd stage was the appearance of larval intermediate stages in the 4th stage. Figures 1–4 are photographs of the different stages. Early-intermediate 4th stage larvae were defined by their close resemblance to normal 3rd stage larvae. They had smaller antennae, carapace, and chelae lengths and molted earlier than the other groups (Table II). The exopodites (swimming appendages) were not greatly reduced as they were in the late-intermediates and normal 4th stage larvae (see below). Dorsal spines were still present and not reduced on the abdomen. Neither the tail spine nor telson lateral spines were reduced. Telson setae were not apparent on the fringe region in the early-intermediates. Their behavior closely resembled that of normal 3rd stage larvae. They were consistently observed to lie in a curled position with the abdomen flexed under the cephalothorax.

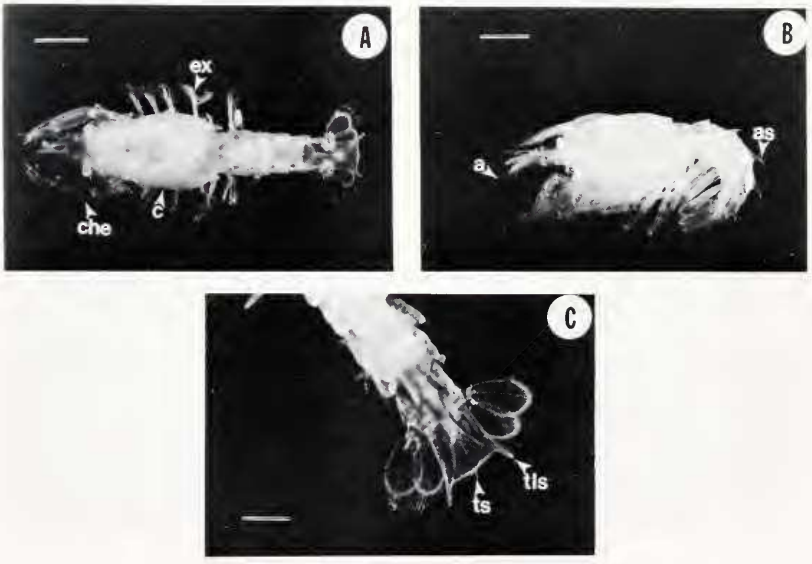

FIGURE 1. Early-intermediate larva at 4th stage. A) Dorsal view showing small chela (che), broad carapace (c), and exopodite (ex). Scale bar = 1 mm. B) Lateral view showing antenna (a) and abdominal spine (as). Scale bar = 1 mm. C) Dorsal view of tail showing telson spine (ts) and telson lateral spine (tls). Scale bar = 0.5 mm.

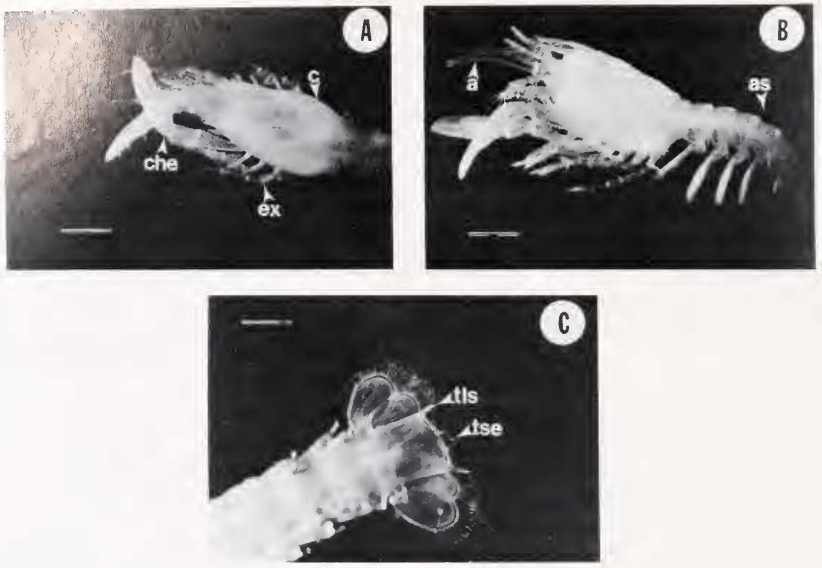

FIGURE 2. Late-intermediate larva at 4th molt. A) Dorsal view showing larger chela (che), normal-shaped carapace (c), and exopodite (ex). Scale bar = 1 mm. B) Lateral view indicating larger antenna (a) and reduced abdominal spine (as). Scale bar = 1 mm. C) Dorsal view of tail showing telson setae (tse) and reduced telson lateral spine (tls). Scale bar = 0.5 mm.

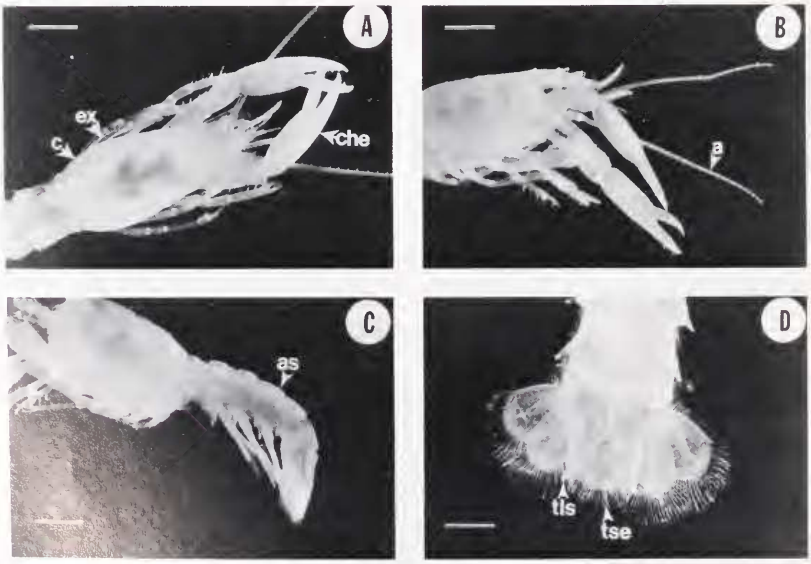

FIGURE 3. Normal 4th stage, eyestalk-ablated larvae. A) Dorsal view indicating chela (che), normal carapace (c), a lack of exopodite (ex). Scale bar = 1 mm. B) Lateral view of cephalothorax region showing normal length antenna (a). Scale bar = 1 mm. C) Lateral view of abdomen showing lack of abdominal spine (as). Scale bar = 1 mm. D) Dorsal view of tail region indicating normally reduced telson lateral spine (tls) and numerous telson setae (tse). Scale bar = 0.5 mm.

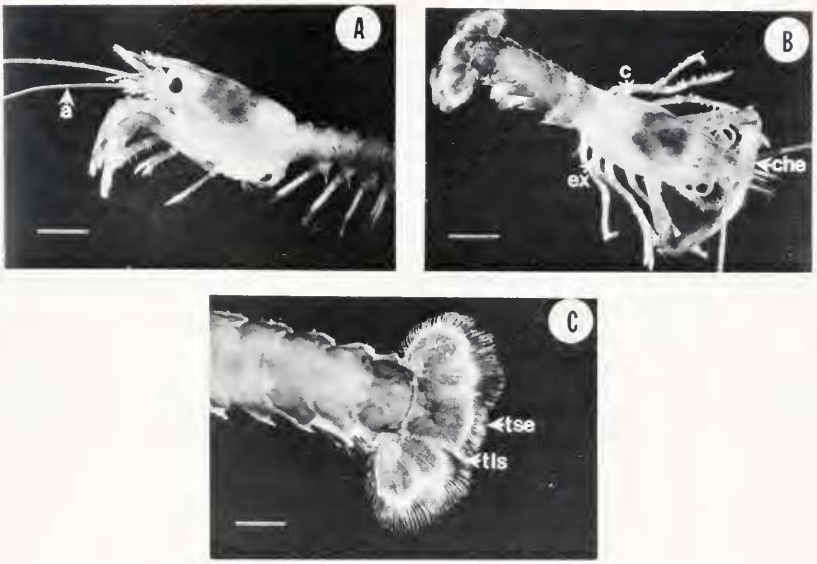

FIGURE 4. Normal 4th stage larvae. A) Lateral view showing antenna (a). Scale bar = 1 mm. B) Dorsal view indicating chela (che), carapace (c), and lack of exopodite (ex). Scale bar = 1 mm. C) Dorsal view of tail indicating reduced telson lateral spine (tls) and numerous telson setae (tse). Scale bar = 0.5 mm.

Normal 4th stage eyestalk-ablated larvae were significantly larger than controls and molted earlier to the 4th stage. Morphological development was equivalent to intact 4th stage larvae except for the size increases displayed by the eyestalk-ablated larvae.

Late-intermediate 4th stage larvae were morphologically more similar to normal (normally appearing) 4th stage larvae than were the early-intermediates. Antennae lengths were smaller than either the normal 4th stage ablated or intact control groups. Chelae lengths were larger than intact controls but smaller than normal 4th stage ablated larvae. Carapace lengths were larger than intact controls and equivalent to

TABLE II

Morphological and 3rd stage molt interval differences between early-intermediate (early-int., n = 102), late-intermediate (late-int., n = 29), normal-ablated (normal-abl., n = 32), and intact (control, n = 60) larval lobsters at 4th stage (means ± S.D.)

Treatment	Antenna length (mm)	Carapace length (mm)	Chela length (mm)	3rd stage molt interval (days)
Early-int.	2.6 ± 0.4	3.9 ± 0.2	2.7 ± 0.2	3.2 ± 0.5
Late-int.	a 4.3 ± 0.6 a	b 4.0 ± 0.1 d	a 3.5 ± 0.2 b	a 3.4 ± 0.5 d
Normal-abl.	6.0 ± 0.5 b	4.1 ± 0.2 a	3.9 ± 0.2 a c	3.8 ± 0.6 c a
Control	5.5 ± 0.5	3.7 ± 0.1	3.3 ± 0.1	4.8 ± 0.7

Statistical significance is indicated by the lines drawn between groups.

[a] Denotes significance at $P < 0.001$, [b] denotes significance at $P < 0.01$, [c] denotes significance at $P < 0.05$, [d] denotes lack of significance.

normal 4th stage ablated larvae (Table II). The exopodites were further reduced compared to the early-intermediates. The tail spine was much reduced or absent in these larvae. Telson lateral spines were more reduced and some fringe setae were apparent, but were not as numerous as those of normal 4th stage ablated larvae. Late-intermediate larvae molted significantly sooner than intact controls, however, not significantly sooner than normal 4th stage ablated larvae.

Intermediate stages appeared to a greater percentage the earlier the time of ablation of those larvae reared at 22°C (Table III). If eyestalks were removed up to 3 h after the molt to 2nd stage, 100% of the larvae molted to early-intermediates. From 6 to 18 h, about 80% became early-intermediates. There was a gradual decrease in the number of early-intermediates and a concomitant increase in late-intermediate and normal 4th stage ablated larvae when eyestalks were removed up to 48 h after the molt to 2nd stage. All of the intact controls molted to normal 4th stage larvae. Larvae from two additional females, however, did not form larval intermediate stages when eyestalk-ablated and reared at 22°C. These intermediate larval stages were rarely observed in many other experiments conducted at 18–20°C.

The effect of rearing conditions on the formation of larval intermediates was also investigated. Table IV shows the results of this experiment (conducted at 18°C). Almost all (97%) of the larvae that were ablated 12 h after molt to 2nd stage, returned to communal rearing kreisels, and fed frozen adult *Artemia,* developed into early-intermediates at the 4th stage. However, when larvae were ablated 90 h after molt to 2nd stage and returned to rearing kreisels, only 17.5% developed into early intermediates. Another 30% molted to late-intermediates and the rest molted to normal 4th stage ablated larvae. None of the kreisel-reared intact controls molted to intermediate stages. Upon molting to 2nd stage, an additional group of larvae from the same hatch were individually transferred from the kreisels to the compartmented trays and fed living *Artemia* nauplii. Some of these were ablated at either 12 or 90 h after the molt to 2nd stage. Ninety-one percent of the 12 h-ablated and 100% of the 90 h-ablated larvae molted to normal 4th stage larvae. As in the kreisels, none of the tray-reared intact controls molted to intermediate stages. Thus, depending upon the rearing conditions, either normal or intermediate larval forms could be induced in destalked larvae.

TABLE III

Morphological characteristics at the 4th stage of lobster larvae ablated at different times

Time of ablation[a] (h)	*n*	4th stage (%)		
		Early-intermediates	Late-intermediates	Normal 4th
1	12	12 (100)	0 (0)	0 (0)
3	14	14 (100)	0 (0)	0 (0)
6	9	6 (67)	2 (22)	1 (11)
12	15	13 (87)	1 (7)	1 (7)
18	16	14 (88)	1 (6)	1 (6)
24	16	8 (50)	3 (19)	5 (31)
30	18	11 (61)	4 (22)	3 (17)
36	17	5 (29)	4 (24)	8 (47)
42	18	6 (33)	7 (39)	5 (28)
48	18	3 (17)	7 (39)	8 (44)
Intact controls	60	0 (0)	0 (0)	60 (100)

[a] Refers to time of ablation following the molt to 2nd stage.

TABLE IV

Morphological characteristics at the 4th stage of eyestalk-ablated and control lobster larvae reared under different conditions

Time of ablation[a] (h)	% Mortality	*n*	4th stage (%) Early-intermediates	Late-intermediates	Normal 4th
12, tray[b]	9.0	33	2 (6.1)	1 (3.0)	30 (90.9)
12, kreisel[c]	33.7	67	65 (97.0)	2 (3.0)	0 (0)
90, tray	16.0	21	0 (0)	0 (0)	21 (100)
90, kreisel	70.0	40	7 (17.5)	12 (30.0)	21 (52.5)
Control, tray	5.3	18	0 (0)	0 (0)	18 (100)
Control, kreisel	15.0	74	0 (0)	0 (0)	74 (100)

[a] Refers to eyestalk ablation at either 12 or 90 h after the molt to 2nd stage. Controls were intact larvae.
[b] Refers to rearing in compartmented trays and fed living *Artemia* nauplii.
[c] Refers to rearing in communal larval systems and fed frozen *Artemia* adults.

DISCUSSION

Molting rates

In general, eyestalk removal induces precocious molting in decapod crustaceans. This includes juvenile (Mauviot and Castell, 1976; Trider *et al.*, 1979; Chang and Bruce, 1980) and adult *Homarus americanus* (Mauviot and Castell, 1976). Our results support the observations of others of molt and growth rate enhancement by destalking lobster larvae (Rao *et al.*, 1973; Charmantier *et al.*, 1985). Removal of eyestalks early in the 2nd stage resulted in a slightly accelerated molt to 3rd stage. The molt intervals of the following stages were more significantly shortened. These results can be compared with work on other decapod larvae. Le Roux (1979) found that the first molt interval was unaffected following ablation in the anomuran *Pisidia longicornis*. The megalopal stage, however, was accelerated. The megalopal molt is accelerated in the crab *Callinectes sapidus* only if eyestalks are removed within 12 h of the previous ecdysis (Costlow, 1963). In a similar study of the crab *Sesarma reticulatum*, Costlow (1966a) reported that molting of magalopae and later stages was accelerated by ablation following the appearance of stalked eyes during zoeal development. Molting rates were unaffected by ablation in either zoeal or megalopal stages of the crab *Rhithropanopeus harrisii* (Costlow, 1966b). However, later work on *R. harrisii* larvae (Freeman and Costlow, 1980) identified an enhanced molting rate in megalopae only when eyestalks were removed within 12 h after the final zoeal molt. Those observations of molt acceleration by destalking of larval crustaceans support the original hypothesis of Brown and Cunningham (1939) on the control of molting by molt-inhibiting hormone.

Larval development

In addition to precocious molting, eyestalk removal can also lead to the development of intermediate stages. Our experiments were designed to examine the effects of rearing conditions on the induction of these intermediate stages in eyestalk-ablated lobster larvae. Eyestalk ablation of early and late 2nd stage larvae, that were reared in recirculating systems and fed frozen *Artemia* (know to be an inferior food source; Sorgeloos, 1980), resulted in numbers of 4th stage early-intermediates equivalent to those reported by Charmantier *et al.* (1985). Alternatively, larvae from the same hatch

were removed, ablated at identical times, and then fed living *Artemia* nauplii (known to be a superior food source; Sorgeloos, 1980) in compartmented culture. These formed <10% intermediates when ablated early in 2nd stage and were 100% normal if ablated to the late premolt period. These results indicate that under suboptimal rearing conditions, eyestalks may not mediate the morphological aspects of metamorphosis.

Usually a finite number of larval stages are described for any given species (reviewed by Williamson, 1982). Stressful conditions can result in extra intermediate stages of natural or laboratory-reared crustacean larvae. However, larval development can be quite variable. Extra larval stages, or the retention of larval characteristics in postlarval stages can be induced by a variety of factors. Temperature (Boyd and Johnson, 1963; Knowlton, 1974), maternal influence (Templeman, 1936), diet (Broad, 1957; Knowlton, 1974; Sulkin, 1975, 1978), and starvation periods (McConaugha, 1982; Anger, 1984) have been identified or suggested as affecting morphological development. There are also a number of accounts of "larval intermediate stages" taken in plankton samples (Pike and Williamson, 1961; Nichols and Keney, 1963, as cited in Costlow, 1965; Knowlton, 1974, for review).

Larval intermediate stages can also result from eyestalk extirpation of decapod larvae. Little (1969) found extra larval stages in the shrimp *Palaemon macrodactylus* as a result of ablation during the 2nd–4th zoeal periods. Our first experiment on time of extirpation showed that nearly 100% of lobster larvae molted to early-intermediates when the operation was performed prior to 18 h of the 2nd stage. A greater percentage of larvae molted to late-intermediates or normal 4th stage larvae as the time of ablation approached 48 h. This period corresponds to early premolt as defined by changes in ecdysteroid titers (Snyder and Chang, 1986). Intermediate stage larvae also completed ecdysis earlier than the controls, although the difference was not statistically significant from late-intermediates. In a study by Le Roux (1979), *P. longicornis* retained some larval characteristics as juveniles when destalked on the 1st day of the 2nd zoeal stage. Costlow (1966b) observed that the crab *S. reticulatum* often went through a "2nd megalops" stage when larvae were destalked as early 2nd zoea. Critical periods for intermediate stage formation during zoeal development of *R. harrisii* (Costlow, 1966a) and *H. americanus* (Charmantier *et al.*, 1985) have also been described. The results reported here support the recurrent theme of a critical early zoeal period when suboptimal factors can result in subsequent intermediate stage formation. This observation is similar to that of Anger and Dawirs (1981) who described such a phenomenon with starvation. Survival of zoea to the next stage depended on their feeding from the beginning to at least 20% of the molt interval.

We observed that eyestalk-ablated larvae occasionally formed intermediates at the 4th stage. These observations imply that the stimulus for molting may take precedence over that for the completion of normal morphological development. This concept is supported by previous observations by others (Costlow, 1966a, b; Knowlton, 1974; McConaugha, 1982). However we were unable to repeat these results with larvae from two other female lobsters. Thus both parental and stress components may affect development (Templeman, 1936; Dawirs, 1983), possibly by the production of larvae that have insufficient energy reserves. Support for the importance of parental factors is the observation that broods from different females require varying lengths of time to complete metamorphosis in the laboratory (Boyd and Johnson, 1963; Costlow, 1965; Rabalais and Cameron, 1985).

The importance of food quality and nutritive reserves in larval development has been illustrated. Broad (1957) demonstrated that both food quality and quantity affected the molting rate and number of larval stages of two shrimp species. There is much evidence to support the idea that a diet of live *Artemia* nauplii or adults is capable of

supporting normal early developmental processes in cultured marine animals (Fluchter, 1980; Sorgeloos, 1980). Normal growth is also supported by *Artemia* which have been preserved by rapid freezing in liquid nitrogen. Slow freezing (the process by which commercially available *Artemia* are frozen) results in a diet that does not support normal development, presumably due to the loss of nutrients (Fluchter, 1980). In our experiments, larvae were destalked after the molt to 2nd stage and fed either commercially frozen *Artemia* adults or freshly hatched nauplii. Virtually all of the ablated larvae that were fed nauplii developed normally to the 4th stage. Those fed commercially frozen adult *Artemia* exhibited delayed morphological development by molting to intermediate stages instead of normal 4th stage ablated larvae. These results extend earlier published observations that food quality may be a factor in the formation of larval lobster intermediate stages.

Eyestalk ablation is known to result in greatly increased respiration rates (Passano, 1961; McWhinnie and Kirchenberg, 1962). Alterations in amino acid (McWhinnie and Mohrherr, 1970), lipid (O'Connor and Gilbert, 1969), and sterol (Spaziani *et al.*, 1982) metabolism result from ablating adult decapods. Amino acid metabolism is also affected by eyestalk extirpation in *Callinectes sapidus* larvae (Tucker and Costlow, 1975). Coupled with the trauma that results from eyestalk ablation, and the inability to visualize prey, the above data support the idea that eyestalk-ablation alters normal life processes to an extent that, without high quality food, normal larval development is not possible.

This situation may be similar to that found in the holometabolous insect, the tobacco hornworm *Manduca sexta* (Safranek and Williams, 1984a, b). Malnourished larvae initiated metamorphosis only after reaching a critical weight. Supernumerary larval stages resulted if molting was initiated prior to the attainment of this weight. Analogous information on the control of metamorphic development in crustacean larvae is lacking.

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LITERATURE CITED

AIKEN, D. E. 1980. Molting and growth. Pp. 91–163 in *The Biology and Management of Lobsters,* Vol. 1, J. S. Cobb and B. F. Phillips, eds. Academic Press, New York.

ANGER, K. 1984. Influence of starvation on moult cycle and morphogenesis of *Hyas araneus* larvae (Decapoda, Majidae). *Helgol. Meeresunters.* **38:** 21–33.

ANGER, K., AND R. R. DAWIRS. 1981. Influence of starvation on the larval development of *Hyas araneus* (Decapoda, Majidae). *Helgol. Meeresunters.* **34:** 287–311.

BOYD, C. M., AND M. W. JOHNSON. 1963. Variations in the larval stages of a decapod crustacean, *Pleuronectes planipes* (Galatheidae). *Biol. Bull.* **124:** 141–152.

BRICK, R. W. 1974. Effects of water quality, antibiotics, phytoplankton, and food on survival and development of larvae of *Scylla serrata* (Crustacea: Portunidae). *Aquaculture* **3:** 231–244.

BROAD, A. C. 1957. The relationship between diet and larval development of *Palaemonetes. Biol. Bull.* **112:** 162–170.

BROWN, F. A. AND O. CUNNINGHAM. 1939. Influence of the sinus gland of crustaceans on normal viability and ecdysis. *Biol. Bull.* **77**: 104–114.

CHANG, E. S., AND M. J. BRUCE. 1980. Ecdysteroid titers of juvenile lobsters following molt induction. *J. Exp. Zool.* **214**: 157–160.

CHANG, E. S., AND D. E. CONKLIN. 1983. Lobster (*Homarus*) hatchery techniques. Pp. 271–275 in *CRC Handbook of Mariculture,* Vol. 1, J. P. McVey, ed. CRC Press, Boca Raton.

CHARMANTIER, G., M. CHARMANTIER-DAURES, AND D. E. AIKEN. 1985. Intervention de pédoncules oculaires dans le contrôle de la métamorphose chez *Homarus americanus* H. Milne-Edwards, 1837 (Crustacea, Decapoda). *C. R. Acad. Sci. Paris* **300**: 271–276.

COSTLOW, J. D. 1963. The effect of eyestalk extirpation on metamorphosis of megalops of the blue crab, *Callinectes sapidus* Rathbun. *Gen. Comp. Endocrinol.* **3**: 120–130.

COSTLOW, J. D. 1965. Variability in larval stages of the blue crab, *Callinectes sapidus. Biol. Bull.* **128**: 58–66.

COSTLOW, J. D. 1966a. The effect of eyestalk extirpation on larval development of the crab, *Sesarma reticulatum* Say. Pp. 209–224 in *Some Contemporary Studies in Marine Science,* H. Barnes, ed. George Allen and Urwin, London.

COSTLOW, J. D. 1966b. The effect of eyestalk extirpation on larval development of the mud crab, *Rhithropanopeus harrisii* (Gould). *Gen. Comp. Endocrinol.* **7**: 255–274.

COSTLOW, J. D. 1968. Metamorphosis in crustaceans. Pp. 3–41 in *Metamorphosis: A Problem in Developmental Biology,* W. Etkin and L. I. Gilbert, eds. Appleton-Century-Croft, New York.

DAVIS, C. C. 1964. A study of the hatching process in aquatic invertebrates. XIII. Events of eclosion in the American lobster, *Homarus americanus* Milne-Edwards (Astacura, Homidae). *Am. Midl. Nat.* **72**: 203–210.

DAWIRS, R. R. 1982. Methodical aspects of rearing decapod larvae, *Pagurus bernhardus* (Paguridae) and *Carcinus maenas* (Portunidae). *Helgol. Meeresunters.* **35**: 439–464.

FISHER, W. S., AND R. T. NELSON. 1977. Therapeutic treatment for epibiotic fouling on Dungeness crab (*Cancer magister*) larvae reared in the laboratory. *J. Fish. Res. Board Can.* **34**: 432–436.

FLUCHTER, J. 1980. Review of the present knowledge of rearing whitefish (Coregonidae) larvae. *Aquaculture* **19**: 191–208.

FREEMAN, J. A., AND J. D. COSTLOW. 1980. The molt cycle and its hormonal control in *Rhithropanopeus harrisii* larvae. *Dev. Biol.* **74**: 479–485.

GRANGER, N. A., AND W. E. BOLLENBACHER. 1981. Hormonal control of insect metamorphosis. Pp. 105–175 in *Metamorphosis: A Problem in Developmental Biology,* 2nd edition, L. I. Gilbert and E. Frieden, eds. Plenum Press, New York.

HARTNOLL, R. G. 1982. Growth. Pp. 111–196 in *The Biology of Crustacea,* Vol. 2, L. G. Abele, ed. Academic Press, New York.

HEDGECOCK, D. 1983. Maturation and spawning techniques for the American lobster, *Homarus americanus.* Pp. 261–270 in *CRC Handbook of Mariculture,* Vol. 1, J. P. McVey, ed. CRC Press, Boca Raton.

KNOWLTON, R. E. 1974. Larval developmental processes and controlling factors in decapod Crustacea, with emphasis on Caridea. *Thalassia Jugoslav.* **10**: 138–158.

LE ROUX, A. 1979. Effects de l'ablation des pédoncules oculaires sur le developpement de *Pisidia longicornis* (Crustacé, Décapode, Anomoure). *Ann. Endocrinol.* **40**: 79–80.

LITTLE, G. 1969. The larval development of the shrimp, *Palaemon macrodactylus* Rathbun, reared in the laboratory, and the effect of eyestalk extirpation on development. *Crustaceana* **17**: 69–87.

MAUVIOT, J. C., AND J. D. CASTELL. 1976. Molt- and growth-enhancing effects of bilateral eyestalk ablation on juvenile and adult American lobsters (*Homarus americanus*). *J. Fish. Res. Board Can.* **33**: 1922–1929.

MCCONAUGHA, J. R. 1982. Regulation of crustacean morphogenesis in larvae of the mud crab, *Rhithropanopeus harrisii. J. Exp. Zool.* **223**: 155–163.

MCWHINNIE, M. A., AND R. J. KIRCHENBERG. 1962. Crayfish hepatopancreas metabolism and the intermolt cycle. *Comp. Biochem. Physiol.* **6**: 117–128.

MCWHINNIE, M. A., AND C. J. MOHRHERR. 1970. Influence of eyestalk factors, intermolt cycle and season upon ^{14}C-leucine incorporation into protein in the crayfish (*Orconectes virilis*). *Comp. Biochem. Physiol.* **34**: 415–437.

NICHOLS, P. R., AND P. M. KENEY. 1963. Crab larvae (*Callinectes*) in plankton collections from cruises of M/V Theodore N. Gill, South Atlantic Coast of the U. S. 1953–1954. U. S. Fish and Wildlife Service, Special Scientific Report Fisheries No. 448. 14 pp.

O'CONNOR, J. D., AND L. I. GILBERT. 1969. Alterations in lipid metabolism associated with premolt activity in a land crab and freshwater crayfish. *Comp. Biochem. Physiol.* **29**: 889–904.

PASSANO, L. M. 1961. The regulation of crustacean metamorphosis. *Am. Zool.* **1**: 89–95.

PIKE, R. B., AND D. I. WILLIAMSON. 1961. The larvae of *Sirontocaris* and related genera (Decapoda, Hippolytidae). *Crustaceana* 2: 187–208.

RABALAIS, N. N., AND J. N. CAMERON. 1985. The effects of factors important in semi-arid environments on the early development of *Uca subcylindrica*. *Biol. Bull.* **168:** 147–160.

RAO, K. R., S. W. FINGERMAN, AND M. FINGERMAN. 1973. Effects of exogenous ecdysones on the molt cycles of fourth and fifth stage American lobsters, *Homarus americanus*. *Comp. Biochem. Physiol.* **44A:** 1105–1120.

SAFRANEK, L., AND C. M. WILLIAMS. 1984a. Critical weights for metamorphosis in the tobacco hornworm, *Manduca sexta*. *Biol. Bull.* **167:** 555–567.

SAFRANEK, L., AND C. M. WILLIAMS. 1984b. Determinants of larval molt induction in the tobacco hornworm, *Manduca sexta*. *Biol. Bull.* **167:** 568–578.

SKINNER, D. M. 1985. Molting and regeneration. Pp. 43–146 in *The Biology of Crustacea,* Vol. 9, D. E. Bliss and L. H. Mantel, eds. Academic Press, New York.

SNYDER, M. J., AND E. S. CHANG. 1986. Effects of sinus gland extracts on larval molting and ecdysteroid titers of the American lobster, *Homarus americanus*. *Biol. Bull.* **170:** 244–254.

SORGELOOS, P. 1980. The use of brine shrimp *Artemia* in aquaculture. Pp. 25–46 in *The Brine Shrimp Artemia,* Vol. 3, G. Persoone, P. Sorgeloos, O. Roels, and E. Jaspers, eds. Universa Press, Wetteren, Belgium.

SORGELOOS, P., E. BOSSUYT, E. LAVINA, M. BAEZA-MESA, AND G. PERSOONE. 1977. Decapsulation of *Artemia* cysts: a simple technique for the improvement of the use of brine shrimp in aquaculture. *Aquaculture* **12:** 311–315.

SPAZIANI, E., L. S. OSTEDGAARD, W. H. VENSEL, AND J. P. HEGMANN. 1982. Effects of eyestalk removal in crabs: relation to normal premolt. *J. Exp. Zool.* **221:** 323–327.

SULKIN, S. D. 1975. The significance of diet in the growth and development of larvae of the blue crab, *Callinectes sapidus* Rathbun, under laboratory conditions. *J. Exp. Mar. Biol. Ecol.* **20:** 119–135.

SULKIN, S. D. 1978. Nutritional requirements during larval development of the portunid crab, *Callinectes sapidus* Rathbun. *J. Exp. Mar. Biol. Ecol.* **34:** 29–41.

TEMPLEMAN, W. 1936. Fourth stage larvae of *Homarus americanus* intermediate in form between third and fourth stages. *J. Biol. Board Can.* **2:** 349–354.

TRIDER, D. J., E. G. MASON, AND J. D. CASTELL. 1979. Survival and growth of juvenile American lobsters (*Homarus americanus*) after eyestalk ablation. *J. Fish. Res. Board Can.* **366:** 93–97.

TUCKER, R. K., AND J. D. COSTLOW. 1975. Free amino acid changes in normal and eyestalkless megalopae larvae of the blue crab, *Callinectes sapidus,* during the course of the molt cycle. *Comp. Biochem. Physiol.* **51A:** 75–78.

WILLIAMSON, D. I. 1982. Larval morphology and diversity. Pp. 43–110 in *The Biology of Crustacea,* Vol. 2, L. G. Abele, ed. Academic Press, New York.